Nigella Sativa Seeds Ameliorate the Hepatotoxicity and Nephrotoxicity of Colchicine Prolonged Use in Albino Rats: a Biochemical and Histopathological Study

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ABSTRACT

Background: Colchicine has been used to treat many diseases for several decades. It is a safe drug if used for a short period of time and under recommended doses. It has a narrow therapeutic index and colchicine overdose is associated with high mortality rate. Objective: The present study was undertaken to investigate the amelioration effect of Nigella sativa seeds against the toxicity that may occur as a result of prolonged use of colchicine using rats as experimental animals. Results: The results revealed that, the administration of N. sativa seeds significantly increased the levels of liver function enzymes (AST, ALT and ALP) as well as the serum levels of creatinine and urea, while decreased the levels of serum albumin. The histopathological examination revealed that prolonged use of colchicine induced hepatotoxicity and nephrotoxicity. The manifestations were hepatocytes necrosis, dilatation of central vein, and degeneration in the form of pyknosis of hepatocytes nuclei, fibrosis, and cytoplasmic vacuolation. Also, N. sativa seeds significantly increased the MDA content and NO when compared with the negative control group. Moreover, the prolonged use of colchicine led to significant increase in the serum levels of liver function enzymes (AST, ALT and ALP) as well as the serum levels of creatinine and urea, while decreased the level of serum albumin. The histopathological examination revealed that prolonged use of colchicine induced hepatotoxicity and nephrotoxicity. Conclusion: It can be concluded that, prolonged use of colchicine may lead to hepatotoxicity and nephrotoxicity. N. sativa seeds ameliorate these toxicity effects, thus N. sativa can be recommended to patients with chronic diseases such as gout which require colchicines for long periods.

INTRODUCTION

Colchicine is an alkaloid from the family of the spindle poisons, which are mainly used to treat and prevent forms of microcrystalline arthritis, such as gout. Colchicine has a narrow therapeutic index, with no clear-cut distinction between nontoxic, toxic, and lethal doses, causing substantial confusion among clinicians. It causes serious systemic effects if ingested in doses that exceed the recommendations. It has been reported that overdose of colchicine is associated with a high mortality rate (Bismuth and Conso, 1977). Earley symptoms usually include gastrointestinal pain; multiorgan failure typically occurs next, alongside metabolic derangements and bone marrow suppression. Death from acute colchicine poisoning is usually due to hemodynamic collapse and cardiac arrhythmias. It has been reported that cardiogenic shock is a possible early dramatic complication in acute colchicine poisoning when associated with co-ingested compounds and which can be potentiated toxicity (Lainé et al., 2012). After ingestion, colchicine is absorbed from the gastrointestinal tract and undergoes deacetylation in liver. The metabolites undergo widespread enterohepatic recirculation before being excreted in bile and faeces. Renal clearance also accounts for 10 - 20% of colchicine removal. If a toxic amount has been ingested and renal function exists larger fractions can be excreted via this route. Increased urinary excretion of colchicine is possible early after ingestion when associated with co-ingested compounds and which can be potentiated toxicity (Lainé et al., 2012). After ingestion, colchicine is absorbed from the gastrointestinal tract and undergoes deacetylation in liver. The metabolites undergo widespread enterohepatic recirculation before being excreted in bile and faeces. Renal clearance also accounts for 10 - 20% of colchicine removal. If a toxic amount has been ingested and renal function exists larger fractions can be excreted via this route. Increased urinary excretion of colchicine is possible early after ingestion when associated with co-ingested compounds and which can be potentiated toxicity (Lainé et al., 2012).
1977) and animal (Baud et al., 1995) immunotherapy antibodies can be successfully used to treat colchicines overdose. However, there are a few limitations on the use of immunotherapy antibodies such as: availability, timeframe between ingestion and admission, dosage issues and high costs of preclinical trials. Recently, extra corporeal life support (ECLS) was successfully used to save a patient who presented with multiple organ failure caused by colchicines poisoning (Jouffroy et al., 2013).

*Nigella sativa* is a plant of Ranunculaceae family that grows spontaneously and widely in several Southern Mediterranean and Middle Eastern countries. The seeds of *Nigella sativa* or its oil have been used for medicinal purposes as a natural remedy for a number of illnesses and conditions such as: bronchial asthma, rheumatism, hypertension, diabetes, inflammation, cough, headache, eczema, fever and influenza (Burits and Bucar, 2000; Ali and Blunden, 2003; Singh et al., 2005). *N. sativa* seeds have over 100 different chemical constituents including both fixed and essential oils, proteins, alkaloids and saponin. The seeds are characterized by a very low degree of toxicity (Ali and Blunden, 2003). Many researcher have reported the antioxidant activity of *N. sativa* (Burits and Bucar, 2000; Ali and Blunden, 2003; Abdel-Wahhab and Aly, 2005). Thus, the present study aimed to investigate the hepatotoxicity and nephrotoxicity that may induce by colchicines prolonged use in rats and the amelioration effect of *N. sativa* seeds against this toxicity by assessing the biochemical and histopathological changes in rat tissues.

**MATERIALS AND METHODS**

*Preparation of colchicine and N. sativa seeds suspensions:*

Cholchicine drug was obtained from E-I-Nasr pharmaceutical chemicals company (ADWIC) Abu-Zaabal Egypt in the tablet form. One tablet contains 500 microgram of active ingredient was dispersed in 5 ml distilled water.

*Nigella sativa* seeds were purchased from a local market, Taif, KSA. One gram of the seeds was added to 100 ml of distilled water, mixed and grinded by a blender. This was made for 5-6 intervals, 60 seconds each time, at room temperature until complete grinding and mixing to obtain a stock solution of crude suspension just before use.

*Animals and experimental design:*

Thirty adult male albino healthy rats weighing (150 ± 20 g) were used for this study. They were housed in air-conditioned, humidity-controlled animal room at College of Medicine, Taif University. Rats had free access to water and food during the experimental period. The rats were caged in three equal groups (each n=10). The first group healthy untreated rats; served as negative control (NCG); each rat received 1ml of distilled water per day orally (via gavage) for 12 weeks. The second group was given colchicine orally (2mg/kg/day); served as positive control group (PCG) (Terkeltaub et al., 2010) and the third group was given 2mg/kg/day of cholicine and 100mg/kg/day of freshly prepared aqueous suspension of *Nigella sativa* seeds powder; served as *N. sativa* treated group (NSG). Oral ad ministration was ach i eved b y gastric gavage. Biochemical tests and histopathological observations were used to investigate the changes in rat’s liver and kidney.

*Ethical considerations:*

All animal experiments were carried out in accordance with the internationally accepted guidelines for the care and use of laboratory animals. Painful procedures were performed under anesthesia to avoid distress and pain. Our standards of animal care and admistration met those required by applicable international laws and regulations.

*Biochemical Measurements:*

*Preparation of Serum and Tissue Homogenate:*

Rats of each group were euthanized at the end of the experimental period under anesthesia by administration of 0.5 cm of ketamine intraperitoneal. The blood samples were obtained from a cardiac puncture using syringe for the determination of serum enzyme levels. Blood samples were put immediately into ice-chilled disposable glass tubes and kept for 30 min. The serum samples were obtained by centrifuging blood samples at 6,000xg for 15 min at 4°C, and enzyme levels were measured in these serum samples. The liver and kidney samples were dissected and put in Petri dishes. After washing with physiological saline (0.9% NaCl), part of these samples was taken for histopathological investigations and the remaining part was kept at -80°C until used. The collected tissues were grinded with liquid nitrogen in a mortar. The grinded tissues (0.5 g) were then homogenized in 2mL 50mM phosphate buffer (pH 7.8) containing 1mM EDTA and 1% PVP. The homogenate was centrifuged under cooling at 15,000xg for 20 m in, and the supernatant was stored at -80°C until used for the determination of catalase (CAT), glutathione peroxidase (GPx), superoxide dismutase (SOD), th e o tal a mount o f glutathione (GSH) and the content of malondialdehyde (MDA) and nitric oxide (NO). Protein content in the crude extracts
was determined according to the Bradford’s method (Bradford, 1976) with bovine serum albumin (BSA) as the standard.

**Enzyme Activity Assays In The Tissues Supernatant:**

CAT activity in the tissues supernatant was measured by following the consumption of H$_2$O$_2$ using the method of Aebi (1984); SOD was assayed by measuring its ability to inhibit the photochemical reduction of nitroblue tetrazolium (NBT) using the method of Beauchamp and Fridovich (1971) and GPx activity was determined according to the method described by Sazuka et al. (1989).

**Determination of lipid peroxidation, Nitric oxide (NO) and glutathione (GSH):**

Lipid peroxidation expressed as malondialdehyde (MDA) formation was assayed colorimetrically in the tissue supernatant according to the method described by Ohkawa et al., (1979). The assay of nitrite/nitrate, as an indirect measure of NO production, was done according to the method described by Green et al., (1982). Tissue glutathione (GSH) levels were determined according to Ellman (1959).

**Liver and Kidney Function Tests:**

Albumin, creatinine, urea and the activities of aspartate aminotransferase (AST), alanine aminotransferase (ALT) and alkaline phosphatase (ALP) enzymes in the serum were measured using laboratory kits obtained from Crescent Diagnostic Company according to the instruction manual.

**Histopathological Assessment:**

The livers and kidneys of rats from the control and treated groups were rapidly dissected out, washed with saline (0.9% NaCl), cut into small pieces and dropped in 10% neutral buffer formalin in which they were kept for appropriate time. After fixation, they were subjected to the normal procedure for paraffin embedding. Sections were cut by rotatory microtome and mounted on glass slides. The sections were stained by Hematoxylin & Eosin, Mallory and Periodic acid Schiff (PAS). The sections were examined using light microscope (Leica DM 1000) at a magnification of 400X (Drury and Wallington, 1980).

**Statistical Analysis:**

Statistical analysis was performed using SPSS version 16. Variability of results was expressed as mean ± SD. The significance of differences between mean values was determined using one way analysis of variance (ANOVA) test.

**Results**

**Biochemical Results:**

Data represented in table (1) indicates that administration of colchicine alone in the second group (PCG) significantly reduced the activity of antioxidant enzymes; catalase (CAT), superoxide dismutase (SOD) and glutathione peroxidase (GPx). Also, significantly reduced the total amount of glutathione (GSH), but increased significantly the malondialdehyde (MDA) content and nitric oxide (NO) in rat liver and kidney tissues when compared with the negative control group (NCG). The administration of *N. sativa* with colchicine in the third group (NTG) prevented the depletion of antioxidant enzymes; CAT, GPx, and SOD, and the total amount of GSH, while inhibited the increase of MDA and NO when compared with the PCG.

Data in table (2) represents the effect of the treatment with colchicine alone or with *N. sativa* on the activity of liver marker enzymes; ALT, AST and ALP and the serum level of albumin, creatinine and urea in the experimental animals. The data showed that administration of colchicine alone (PCG) significantly increased the activity of ALT, AST and ALP enzymes, as well as the serum levels of creatinine and urea, while decreased the level of albumin when compared with the negative control group (NCG). The administration of *N. sativa* with colchicine in group three (NTG) significantly reduced the activity of ALT, AST and ALP enzymes, as well as the levels of creatinine and urea, while increased the level of serum albumin when compared with the positive control group (PCG) and returned the values of the liver and kidney function tests around the normal control values (NCG).
Histopathological Findings:

Haematoxyline and Eosin stained liver sections are shown in Figure (1). Control liver sections (NCG) showed a normal hepatic structure as normal hexagonal hepatic lobules with portal triads at the vertices and a central vein (Fig 1a). The liver sections of the second group (administered with colchicine alone; PCG) showed marked hepatic disorganization which represented as necrosis of hepatocytes, contracted and fragmented pyknotic nuclei, increased number of cytoplasm vacuoles, degenerated kupffer cells and fibrosis of central vein (Fig 1b). While the liver section of the third group administered with colchicines and N. sativa (NTG) showed some protective effects when compared to the PCG as marked diminution of hydropic degeneration, normal size of central vein and blood sinusoids (Fig 1c). Collagen contents in liver sections stained with Mallory are shown in Figure (2). Normal distribution of wavy fibrils were observed in the liver of NCG (Fig 2a), while in the PCG the wavy collagen fibrils were seen as sporadic fibrils or were fused together to form thick bundles of collagen fibers (Fig 2b). An improvement in the collagen deposition and connective tissue fibers was observed in the NTG as compared to the PCG (Fig 2c).

Table 1: Effect of the treatment with colchicine alone or with Nigella sativa seeds on liver and kidneys antioxidant enzymes, and the contents of GSH, MDA and NO

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Groups</th>
<th>NCG</th>
<th>PCG</th>
<th>NST</th>
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</thead>
<tbody>
<tr>
<td><strong>Liver</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>CAT</td>
<td>36.53 ± 3.15</td>
<td>24.23 ± 5.05*</td>
<td>33.99 ± 6.13*</td>
<td></td>
</tr>
<tr>
<td>SOD</td>
<td>20.01 ± 2.22</td>
<td>13.87 ± 2.21*</td>
<td>18.95 ± 1.71*</td>
<td></td>
</tr>
<tr>
<td>GPx</td>
<td>5.07 ± 0.48</td>
<td>2.32 ± 0.80*</td>
<td>3.99 ± 0.65*</td>
<td></td>
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<tr>
<td>GSH</td>
<td>91.41 ± 5.61</td>
<td>48.23 ± 5.72*</td>
<td>85.11 ± 6.27*</td>
<td></td>
</tr>
<tr>
<td>MDA</td>
<td>22.98 ± 1.35</td>
<td>36.05 ± 1.74*</td>
<td>25.69 ± 1.95*</td>
<td></td>
</tr>
<tr>
<td>NO</td>
<td>87.66 ± 4.47</td>
<td>94.32 ± 7.84*</td>
<td>86.38 ± 6.18*</td>
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<tr>
<td><strong>Kidney</strong></td>
<td></td>
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<td></td>
<td></td>
</tr>
<tr>
<td>CAT</td>
<td>13.53 ± 2.15</td>
<td>8.23 ± 1.05*</td>
<td>12.89 ± 2.13*</td>
<td></td>
</tr>
<tr>
<td>SOD</td>
<td>7.71 ± 1.22</td>
<td>5.87 ± 1.29*</td>
<td>6.75 ± 1.71*</td>
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<tr>
<td>GPx</td>
<td>5.95 ± 0.68</td>
<td>2.75 ± 0.87*</td>
<td>4.49 ± 0.65*</td>
<td></td>
</tr>
<tr>
<td>GSH</td>
<td>49.41 ± 4.51</td>
<td>28.98 ± 6.72*</td>
<td>42.87 ± 5.77*</td>
<td></td>
</tr>
<tr>
<td>MDA</td>
<td>18.98 ± 1.35</td>
<td>26.05 ± 2.95*</td>
<td>19.60 ± 3.05*</td>
<td></td>
</tr>
<tr>
<td>NO</td>
<td>47.66 ± 5.44</td>
<td>54.32 ± 5.91*</td>
<td>49.39 ± 7.58*</td>
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</tbody>
</table>

Values are expressed as mean ± S.D. for ten animals in each group. Colchicine group (PCG) was compared with negative control (NCG) group. N. sativa treated group (NTG) was compared with colchicine group. CAT, nmol of H$_2$O$_2$ consumed/min/mg protein; SOD, units/mg protein (one unit of the SOD activity is the amount of enzyme required to give 50% inhibition of epinephrine auto oxidation); GPx, nmol GSH oxidized/min/mg protein; GSH, μg of GSH/mg protein; MDA, nmol of MDA/mg protein; NO, nmol/mg protein. * p<0.05.

Table 2: Effect of the treatment with colchicine alone or with Nigella sativa seeds on liver and kidneys function tests

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Groups</th>
<th>NCG</th>
<th>PCG</th>
<th>NST</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Liver</strong></td>
<td></td>
<td></td>
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<tr>
<td>AST (U/L)</td>
<td>70.80 ± 8.15</td>
<td>135.8 ± 9.05*</td>
<td>80.19 ± 9.13*</td>
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<tr>
<td>ALT (U/L)</td>
<td>28.71 ± 4.22</td>
<td>48.87 ± 8.21*</td>
<td>32.60 ± 3.71*</td>
<td></td>
</tr>
<tr>
<td>ALP (U/L)</td>
<td>110.77 ± 7.68</td>
<td>159 ± 8.83*</td>
<td>123.59 ± 6.65*</td>
<td></td>
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<tr>
<td><strong>Kidney</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>creatinine (mg/dL)</td>
<td>0.75 ± 0.04</td>
<td>1.07 ± 0.07*</td>
<td>0.87 ± 0.02*</td>
<td></td>
</tr>
<tr>
<td>Urea (mg/dL)</td>
<td>25.4 ± 1.24</td>
<td>41.97 ± 2.57*</td>
<td>28.3 ± 3.04*</td>
<td></td>
</tr>
</tbody>
</table>

Values are expressed as mean ± S.D. for ten animals in each group. Colchicine group (PCG) was compared with negative control (NCG) group. N. sativa treated group (NTG) was compared with colchicine group. * p<0.05.
Figure 3 illustrates the Periodic Acid Schiff’s (PAS) stained liver sections. The liver sections of NC showed mucopolysaccharide granules in the cytoplasm of hepatocytes; the peripheral zonal, cell is showed a higher mucopolysaccharide content than the central zonal cells (Fig. 3a). While the sections of PCG liver sections showed severe reduction in the total amount of PAS positive material (Fig. 3b). The NTG liver slides showed a mild reduction of PAS positive material but did not reach to the level of the control group (Fig. 3c).
Kidney sections stained with Haematoxyline and Eosin are shown in Figure (4). Control Kidney sections (NCG) showed normal renal structure (Fig 4a), while the sections of PCG kidney tissues showed severe tubular damage, enlarged vascular glomeruli, tight filling of Bowman’s capsule and absence of capsular spaces (Fig 4b). The kidney sections of the third group rats (NTG) showed normal renal glomeruli, normal proximal and distal convoluted tubules (Fig 4c). Collagen contents in Kidney tissues stained with Mallory are shown in Figure (5). Normal renal structure and normal distribution of collagen were observed in The Kidney tissues of control rat (Fig 5a). The PCG treated with colchicine showed fibrosis of vascular glomeruli, degenerated epithelial lining Bowman’s capsule with oedema and fibrosis of tubular epithelium cells (Fig 5b), whereas the kidney sections of NTG showed improvement in the collagen deposition and connective tissue fibers when compared to the kidney of the PCG (Fig 5c). Normal positive reaction with PAS was observed in the Kidney tissues of NCG (Fig 6a), while the PCG showed marked decrease in positivity of PAS reaction (Fig 6b). The NTG showed an increase in the positivity of PAS reaction (Fig 6c).

Fig. 4: Photomicrography of kidney tissues stained with H&E: a) Light photomicrography of kidney tissue of a control rat (NCG) the renal glomeruli (G) has flat epithelium lining the glomerular capsule (BC) with distinct capsular space (CP), normal proximal and distal convoluted tubules (T). b) The kidney tissue of the second group (PCG) rats showing decrease of renal glomeruli vasculature (G) tight filling of glomerular capsular space (CP), with degenerated epithelial lining the Bowman’s capsule (BC), oedema and degeneration of some tubular epithelium cells (T). c) The kidney tissue of the third group (NTG) showing normal glomeruli (G), capsular space (CP), with normal proximal and distal convoluted tubules (T). X400

Fig. 5: Photomicrography of kidney tissues stained with Mallory: a) the kidney tissues of the control rat (NCG) showing normal glomeruli (G) and flat epithelium lining glomerular capsule (BC) with distinct capsular space (CP) with normal proximal and distal convoluted tubules (T). b) The kidney tissues of rat from the second group (PCG) showing fibrosis of vascular glomeruli (G), tight filling of glomerular capsular space (CP), with degenerated epithelial lining Bowman’s capsule (BC), oedema and fibrosis of tubular epithelium cells (T). c) The kidney tissues of rat from the third group (NTG) showing normal glomeruli (G), glomerular capsule (BC) normal capsular space (CP) with normal proximal and distal convoluted tubules (T). X400
Discussion:
Colchicine is a safe drug if used for a short period of time and under recommended doses. In the cases of chronic diseases such as gout and familial Mediterranean fever, patients have to use it for prolonged periods. It has been reported that overdose of colchicine is associated with a high mortality rate (Bismuth and Conso, 1977). Therefore, in the present study we aimed to investigate the amelioration effect of *N. sativa* seeds against the toxicity that may occur as a result of prolonged use of colchicine using rats as experimental animals. The main organs that involve in its metabolism and excretion (liver and kidney) were chosen for this biochemical and histopathological study to assess the changes that may arise as consequences of colchicine prolonged use. The choice of *N. sativa* was based on its antioxidant activity that has been reported by many researchers (Burits and Bucar, 2000; Ali and Blunden, 2003; Abdel-Wahhab and Aly, 2005), in addition to its very low degree of toxicity (Ali and Blunden, 2003).

Antioxidant enzymes such as CAT, SOD and GPx and preventive antioxidants such as glutathione (GSH) are the first line of defense against ROS. Oxidative stress occurs when the balance between reactive oxygen species (ROS) generating systems and antioxidative systems is disrupted. Overproduction of ROS leads to lipid peroxidation, protein oxidation and DNA oxidation. It has been reported that colchicine has a potential effect on increased protein oxidation and lipid peroxidation, owing to the overproduction of reactive oxygen species (ROS) and decreased efficiency of antioxidant defenses (Gulbahar et al., 2007). Therefore, in the current study oxidative stress biomarkers malondialdehyde (MDA) and nitric oxide (NO) levels, as well as the antioxidant content (GSH) and the activities of CAT, SOD and GPx antioxidant enzymes were assayed in rat’s liver and kidney tissues supernatant to explore the toxicity effects of colchicine and the modulation effect of *N. sativa* seeds against this toxicity.

The biochemical results shown in Table (1) revealed that the administration of colchicine significantly reduced the activity of antioxidant enzymes: CAT, SOD and GPx and reduced the total amount of GSH. On the other hand, colchicine significantly increased the MDA content and NO in rat liver and kidney tissues when compared with the negative control group (NCG). It is clear from these results that colchicine disrupted the balance between ROS generating systems and antioxidant agents, which led to overproduction of ROS in rat tissues. The overproduction of ROS led to lipid peroxidation indicated by an increase in MDA content, which in turn led to hepatic and kidney dysfunction and damage. These results were in agreement with Gulbahar et al. (2007). The administration of *N. sativa* with colchicine (NTG) prevented the depletion of antioxidant enzymes: CAT, SOD, and GPx, and the total GSH, and inhibited the increase of MDA and NO when compared with the PCG. These results reveal the antioxidant capacity of *N. sativa* as reported by many researchers (Burits and Bucar, 2000; Ali and Blunden, 2003; Abdel-Wahhab and Aly, 2005).

It is known that several biochemical tests are useful in the evaluation of hepatic and kidney dysfunction and damage. For example: some of liver function tests are associated with functionality (e.g., albumin); some with cellular integrity [e.g., transaminase (AST&ALT)] and some with conditions linked to the biliary tract [e.g., alkaline phosphatase (ALP) and gamma-glutamyl transferase (GGT)]. Albumin levels are decreased in chronic liver disease, such as cirrhosis. It is also decreased in nephrotic syndrome, where it is lost through the urine. AST is similar to ALT in that it is another enzyme associated with liver parenchymal cells. These enzymes level rise in acute liver damage. ALP is an enzyme in the cells lining the biliary ducts of the liver. ALP level in plasma is with large bile duct obstruction, intrahepatic cholestasis or infiltrative diseases of the liver. Furthermore, renal function is an indication of the state of the kidney and its role in renal physiology (Sallie et al., 1991; Levey et al., 2003; Rosenberg et al., 2004; McPhee and Papadakis, 2010). The result of the current study revealed that the prolonged use of colchicine led to significant increase in the serum level of AST, ALT,

**Fig. 6:** Photomicrography of kidney tissues stained with Periodic acid-Schiff’s: a) control rat kidney showing normal positive reaction of PAS. b) The kidney tissues of rat from the second group (PCG) showing marked decrease in PAS reaction increase number of vacuoles (v). c) The kidney tissues of rat from the third group (NTG) showing return normal positive reaction of PAS. Normal glomeruli (G) and blood sinusoids (BS). X400
ALP, creatinine and urea, while decreased the levels of serum albumin. These results were consistent with the histopathological observations, which indicated the harmful effects of colchicine prolonged use on the rat’s liver and kidney. Our results in agreement with many studies demonstrated the detrimental effects of long-term use of colchicine (Ehrenfeld et al., 1987; Kuncl et al., 1987; Crocenzi et al., 1997; Masuda et al., 1998; Zemer et al., 1991). On the other hand, the administration of N. sativa with colchicine (NTG) reduced its toxicity indicating the amelioration effects of N. sativa. Our results are in agreement with many researchers (Al-Ghamdi, 2003; Ali and Blunden, 2003; Kanter et al., 2005). It has been reported that the pharmacological actions of the crude extracts of the seeds include protection against nephrotoxicity and hepatotoxicity induced by either disease or chemicals. It would appear that the beneficial effects of the use of the seeds and thymoquinone might be related to their cytoprotective and antioxidant actions, and to their effect on some mediators of inflammation (Ali and Blunden, 2003).

**Conclusion:**
Together, the biochemical and histopathological findings of the current study clearly indicated that prolonged use of colchicine has harmful effects on liver and kidney and long-term use may lead to hepatotoxicity and nephrotoxicity. The administration of N. sativa with colchicines ameliorated these toxicity effects.

**Recommendation:**
We can recommend the consumption of N. sativa as a safe preventive agent against the colchicine toxicity to patients with chronic diseases such as gout which requires treatment with colchicine for long periods.

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